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EXAMINER
KERR, KATHLEEN M

ART UNIT	PAPER NUMBER
1652	13

DATE MAILED: 08/01/2003

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application N .	Applicant(s)	
	09/767,680	GEPPEL ET AL.	
	Examin r	Art Unit	
	Kathleen M Kerr	1652	

-- The MAILING DATE of this communication appears on the c ver sheet with th c rrespondenc address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *30 May 2003* .

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1 and 4-55* is/are pending in the application.

4a) Of the above claim(s) *18-33 and 53-55* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1,4-17 and 34-52* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☒ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

11)☐ The proposed drawing correction filed on _____ is: a)☐ approved b)☐ disapproved by the Examiner.

If approved, corrected drawings are required in reply to this Office action.

12)☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. §§ 119 and 120

13)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____ .

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

14)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application).

a) ☐ The translation of the foreign language provisional application has been received.

15)☒ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO-1449) Paper No(s) _____ .
4) ☐ Interview Summary (PTO-413) Paper No(s). _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other:

DETAILED ACTION

Application Status

1. In response to the previous Office action, a non-Final rejection (Paper No. 10, mailed on December 31, 2002), Applicants filed a response and amendment received on May 30, 2003 (Paper No. 12). Said amendment cancelled Claims 2 and 3, amended Claims 1, 4, 5, 7, 8, 10, 13, 15, 16, and 17, and added new Claims 34-55. Thus, Claims 1 and 4-55 are pending in the instant Office action.

Restriction/Election

2. Newly filed claims 34-55 are not all drawn to the elected invention. Applicants previously elected cell and starter culture compositions thereof, Claim 1-17. New Claims 34-52 are drawn to the elected invention. However, new Claims 53-55 are drawn to subject matter of previously identified Groups III and IV, and thus, are withdrawn from further consideration.

Claims 1 and 4-55 are pending in the instant application. Claims 1, 4-17, and 34-52 are drawn to the elected invention and will be examined herein. Claims 18-33 and 53-55 are withdrawn from further consideration as non-elected inventions.

Priority

3. As previously noted, the instant application is granted the benefit of priority as the continuation of PCT/DK01/00036 filed on January 18, 2001 and as a continuation-in-part of U.S. non-provisional Application No. 09/488,644 filed on January 21, 2000. The previous statement

about the priority of Claim 5 (cytochrome d) is withdrawn by virtue of Applicants pointing out clear support for the claim in the priority document.

Withdrawn - Objections to the Specification

4. Previous objection to the specification for lacking updated continuity data in the first paragraph is withdrawn by virtue of Applicants' amendment.

5. Previous objection to the specification for not labeling the description of the drawings is withdrawn by virtue of Applicant's amendment.

6. Previous objection to the Abstract for not completely describing the disclosed subject matter is withdrawn by virtue of Applicants' amendment.

Withdrawn - Claim Objections

7. Previous objection to Claim 5 for having improper form is withdrawn by virtue of Applicants' amendment.

8. Previous objection to Claim 13 under 37 C.F.R. § 1.75(c), as being of improper dependent form for failing to further limit the subject matter of a previous claim is withdrawn by virtue of the Examiner's reconsideration. While Claim 1 can be drawn to, for example, freeze dried cells, Claim 13 requires a culture medium. As such, it does effectively further limit Claim 1.

Withdrawn - Claim Rejections - 35 U.S.C. § 112, second paragraph

9. Previous rejection of Claims 1-17 under 35 U.S.C. § 112, second paragraph, as being indefinite for the term "culturally modified" is withdrawn by virtue of Applicants' amendment. The Examiner will, however, expound on arguments presented by Applicants for clarity of the record.

Applicants traverse the holding that "a specific genetic alteration is likely to explain the effect discovered by Applicants". The Examiner was under the impression that the addition of porphyrin to a culture somehow altered the cells such that when cultured again in the absence of porphyrin, a lasting effect is seen. This is not the case; or, at least, this case is not described in the specification. The examples in the instant specification are limited to the treatment of *L. lactis* cell culture with 10 mg/L haemin. Cells are then harvested and frozen. These cells are lysed and assayed for NOX and LDH activity. Whole cells also are also added to milk for 2 hours at 30°C (a fermentation, but limited) to measure their ability to remove O_2 from the milk (pasteurization). Similar cells are also lysed and assayed for their cytochrome content. At no time are the cells re-cultured having some long-term, permanent effect.

As written, the claim is a product by process claim, acceptable to Office policy.

10. Previous rejection of Claims 3-5 under 35 U.S.C. § 112, second paragraph, as being indefinite for the term "detectable amount" is withdrawn by virtue of Applicants' amendment.

11. Previous rejection of Claims 7 and 17 under 35 U.S.C. § 112, second paragraph, as being indefinite for the term "including" is withdrawn by virtue of Applicants' amendment.

12. Previous rejection of Claims 10-12 under 35 U.S.C. § 112, second paragraph, as being indefinite for the abbreviations "NOX" and "LDH" is withdrawn by virtue of Applicants' amendment.

Maintained - Claim Rejections - 35 U.S.C. § 112, second paragraph

13. Previous rejection of Claims 8 and 9 under 35 U.S.C. § 112, second paragraph, as being indefinite for the term "about" is maintained. Applicants' arguments have been fully considered but are not deemed persuasive. Applicants argue that the term "is commonly used in patent law and it adds breadth to a limitation which it modifies;" this argument is not persuasive without particular citation in "patent law" so that the Examiner can assess its proposed use. Applicants also argue that the scope is expanded by two degrees; however, the Examiner cannot read limitations into the claims as Applicants request. For the scope of the term to be clear, the claim itself must be clear.

Withdrawn - Claim Rejections - 35 U.S.C. § 112, first paragraph

14. Previous rejection of Claims 1-17 under 35 U.S.C. § 112, first paragraph, written description, is withdrawn by virtue of Applicants' amendment and the Examiner's reconsideration. The amended claim structure is a product by process claim. As such, the structure is wholly described by the process (treating with a porphyrin-containing compound) and the starting material (lactic acid bacteria).

15. Previous rejection of Claim 7 is rejected under 35 U.S.C. § 112, first paragraph, enabling deposit, is withdrawn by virtue of Applicants' amendment.

Withdrawn - Claim Rejections - 35 U.S.C. § 102

16. Previous rejection of Claims 1, 6, 7, 10, 11, 13, 14, 15, and 17 under 35 U.S.C. § 102(b) as being anticipated by Kaneko *et al.* is withdrawn by virtue of Applicants' amendment.

17. Previous rejection of Claims 2-5, 8, 9, and 12 under 35 U.S.C. § 102(b) as being anticipated by Kaneko *et al.* as evidenced by Geppel *et al.* is withdrawn by virtue of Applicants' amendment.

NEW ISSUES

Objections to the Specification

18. The specification is objected to for being confusing concerning porphyrin content throughout the examples presented. Specifically, examples detecting haemin in the supernatant and pellet of 10 mg/L haemin treated cells describe that no porphyrin compound (haemin) is found in the supernatant while 41 ppm porphyrin compound (haemin) is found in the cellular pellet. In other words, the specification describes this example as evidence for retention of *generic* porphyrin-containing compounds in the treated cell pellets, while no retention of *generic* porphyrin-containing compounds is identified in the supernatant. This retention in the cell has useful effects in the treated cells (e.g., effect on oxygen consumption). However, in later examples with identically treated cells, porphyrin compound (cytochrome d) is identified in the supernatant at 13 ppm. Thus, it is confusing how the first analysis identifies no porphyrin compound (haemin) in the supernatant while later analysis does find porphyrin (cytochrome d) in the supernatant. Specifically, the use of haemin and/or cytochrome d assays to support generic

claims to retention of any porphyrin compounds is confusing since the results described in the specification are inconsistent. Clarification is required.

Objections to the Claims

19. Claim 14 is objected to for having improper punctuation. The comma after frozen is inappropriate. Correction is required.

Claim Rejections - 35 U.S.C. § 112

The following is a quotation of the second paragraph of 35 U.S.C. § 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

20. Claim 34 is rejected under 35 U.S.C. § 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. It is unclear if DSM12015 must be subjected to the porphyrin-containing substrate treatment or if DSM12015 already has the characteristics claimed in Claim 1. The term "bacterial species" does not have proper antecedent basis in Claim 1. Clarification is required.

The following is a quotation of the first paragraph of 35 U.S.C. § 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

21. Claim 34 is rejected under 35 U.S.C. § 112, first paragraph, enabling deposit, as containing subject matter which was not described in the specification in such a way as to enable

one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention. To use the claimed product, one of skill in the art is required to have DSM 12015. While the instant specification contains deposit information on page 18 and added to the end of the specification, the requirements to enable such a deposit have not been fully met by the instant application. To enable the instant claims by enabling the deposit of DSM 12015, the record must also contain a statement certifying that all restrictions on accessibility to said deposit be irrevocably removed by Applicant upon the granting of the patent (see M.P.E.P. § 2404.01); this statement may be certified by Applicants or Applicants' representative.

This rejection was previously presented against Claim 7. Applicants argued that a Declaration of Deposit was filed along with the amendment/response received. This is not the case; the file contains no such declaration.

22. Claims 1, 4-17, 34-52 are rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, because the specification, while being enabling for lactic acid bacterial cells modified to contain at least 0.1 ppm haemin, does not reasonably provide enablement for lactic acid bacterial cells modified to contain at least 0.1 ppm of ***any*** **porphyrin** containing compound. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make the invention commensurate in scope with these claims. To produce such cells to the full extent of their scope would require undue experimentation.

The factors to be considered in determining whether undue experimentation is required are summarized In re Wands 858 F.2d 731, 8 USPQ2nd 1400 (Fed. Cir, 1988). The Court in Wands states: "Enablement is not precluded by the necessity for some experimentation such as routine screening. However, experimentation needed to practice the invention must not be undue

experimentation. The key word is 'undue,' not 'experimentation.' " (Wands, 8 USPQ2d 1404). Clearly, enablement of a claimed invention cannot be predicated on the basis of quantity of experimentation required to make or use the invention. "Whether undue experimentation is needed is not a single, simple factual determination, but rather is a conclusion reached by weighing many factual considerations." (Wands, 8 USPQ2d 1404). The factors to be considered in determining whether undue experimentation is required include: (1) the quantity of experimentation necessary, (2) the amount or direction or guidance presented, (3) the presence or absence of working examples, (4) the nature of the invention, (5) the state of the prior art, (6) the relative skill of those in the art, (7) the predictability or unpredictability of the art, and (8) the breadth of the claims. While all of these factors are considered, a sufficient amount for a *prima facie* case is discussed below.

In the instant application, *L. lactis* CHCC373 cells are treated with 10 mg/L haemin and grown in anaerobic or aerobic conditions. The cell extract (supernatant) and pellet (debris) were assayed. The assay described shows 41 ppm haemin found in the cell debris, i.e. associated with the cellular material, and not in the cell extract when cells are grown under anaerobic or aerobic conditions. Thus, 10 mg/L haemin-treated *L. lactis* cells retain 41 ppm haemin attached to their cellular material when cells are fermented under aerobic or anaerobic conditions. While minimal guidance in the use of other porphyrin-containing compounds is offered, no other working examples to help define the scope of the claims are described. The state of the prior art is such that numerous porphyrin compounds are known, but all have different cellular association characteristics with different bacteria. Therefore, it is wholly unpredictable how to achieve the

containment of at least 0.1 ppm of *any* porphyrin-containing compound other than those demonstrated, i.e., haemin, in the specification and/or the art.

23. Claim 5 is rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, because the specification, while being enabling for lactic acid bacterial cells modified to contain at least 0.1 ppm **cytochrome d** when fermented under aerobic conditions, does not reasonably provide enablement for lactic acid bacterial cells modified when fermented under **anaerobic** conditions. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make the invention commensurate in scope with these claims. To produce such cells to the full extent of their scope would require undue experimentation.

The factors to be considered in determining whether undue experimentation is required are summarized above.

In the instant application, cells of a mixed strain of *Lactococcus lactis* are treated with 10 mg/L haemin and grown under anaerobic or aerobic conditions. The cell extract (supernatant) was assayed. The assay performed identified cytochrome d in only the aerobically grown cells (not anaerobically grown) and estimates cytochrome d to be present at a concentration of 13 ppm. Thus, 10 mg/L haemin-treated *L. lactis* mixed cells produce and retain 13 ppm cytochrome d in their cells when cells are fermented under aerobic conditions. No guidance as how to achieve these levels of cytochrome d in anaerobically treated cells is offered. No working examples to help define the scope of the claims are described. The state of the prior art is such that numerous porphyrin compounds are known, but all have different levels of cytochrome d with different responses to haemin addition under anaerobic conditions. Therefore, it is wholly

unpredictable how to achieve at least 0.1 ppm of cytochrome d in haemin treated cells under anaerobic conditions.

24. Claims 8-9 are rejected under 35 U.S.C. § 112, first paragraph, enablement, as failing to comply with the enablement requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention. To make the cells claimed that will be effective when **inoculated** in a **concentration** of **10^7 cells/ml** into low pasteurised skimmed milk having **8 ppm of dissolved oxygen** would require undue experimentation.

The factors to be considered in determining whether undue experimentation is required are summarized above.

In the instant application, *L. lactis* CHCC373 cells are treated with 10 mg/L haemin and grown in anaerobic or aerobic conditions. Whole cells where then used to inoculate milk whose initial oxygen concentration was **8.3 mg/kg**; **no inoculation concentration** was described but the fermentation was at 30°C for 2 hours. Anaerobic-treated cells removed 41% and 43% (+/- 10 mg/L haemin) of the dissolved oxygen in 2 hrs; aerobic-treated cells removed 57% and 69% (+/- 10 mg/L haemin) of the dissolved oxygen in 2 hrs. Thus, the experiment described does not support the claim to initial 8 ppm dissolved oxygen and at the particular inoculation concentration of 10^7 cells/ml since no cells are described having 8 ppm O_2 and no inoculation at the claimed level is described. While minimal guidance in the use of other porphyrin-containing compounds is offered, no other working examples to help define the scope of the claims are described. The state of the prior art is such that numerous porphyrin compounds are known, but

all have different oxygen consumption characteristics with different bacteria. Therefore, it is wholly unpredictable how to achieve a cell that consumes at least 25% of the dissolved oxygen when the beginning oxygen level is only 8ppm with the particularly claimed inoculation concentration.

25. Claim 9 is rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, because the specification, while being enabling for lactic acid bacterial cells modified to consume **at least 50% of dissolved oxygen** with treatment under aerobic conditions, does not reasonably provide enablement for lactic acid bacterial cells modified when fermented under **anaerobic** conditions. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make the invention commensurate in scope with these claims. To produce such cells to the full extent of their scope would require undue experimentation.

The factors to be considered in determining whether undue experimentation is required are summarized above.

In the instant application, *L. lactis* CHCC373 cells are treated with 10 mg/L haemin and grown in anaerobic or aerobic conditions. Whole cells where then used to inoculate milk whose initial oxygen concentration was 8.3 mg/kg; no inoculation concentration was described but the fermentation was at 30°C for 2 hours. Anaerobic cells removed 41% and 43% (+/- 10 mg/L haemin) of the dissolved oxygen in 2 hrs; aerobic cells removed 57% and 69% (+/- 10 mg/L haemin) of the dissolved oxygen in 2 hrs. Thus, the experiment described does not support the claim to cells grown anaerobically that can consume 50% of the dissolved oxygen in 2 hours since aerobic conditions are required to produce such a cell. While minimal guidance in the use of other porphyrin-containing compounds is offered, no other working examples to help define

the scope of the claims are described. The state of the prior art is such that numerous porphyrin compounds are known, but all have different oxygen consumption characteristics with different bacteria under different aeration conditions. Therefore, it is wholly unpredictable how to achieve a cell that consumes at least 50% of the dissolved oxygen in 2 hours when the cell was treated under anaerobic conditions with haemin.

26. Claim 11 is rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, because the specification, while being enabling for lactic acid bacterial cells modified to reduce **NOX** activity by **at least 10%** with treatment under aerobic conditions, does not reasonably provide enablement for lactic acid bacterial cells modified when fermented under **anaerobic** conditions. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make the invention commensurate in scope with these claims. To produce such cells to the full extent of their scope would require undue experimentation.

The factors to be considered in determining whether undue experimentation is required are summarized above.

In the instant application, *L. lactis* CHCC373 cells are treated with 10 mg/L haemin and grown in anaerobic or aerobic conditions. The cell extract (supernatant) and pellet (debris) were assayed. The assay performed shows 27% reduced NOX activity with haemin treatment under aerobic conditions in the cell extract, i.e. associated with the cellular material, and no reduction with haemin treatment under anaerobic conditions in the cell extract. Thus, the experiment described does not support the claim to cells grown anaerobically that can reduce NOX activity by at least 10%. While minimal guidance in the use of other porphyrin-containing compounds is offered, no other working examples to help define the scope of the claims are described. The

state of the prior art is such that numerous porphyrin compounds are known, but all have different NOX activity characteristics with different bacteria under different aeration conditions. Therefore, it is wholly unpredictable how to achieve a cell that reduces NOX activity by at least 10% when cells have undergone treatment with haemin under anaerobic conditions.

27. Claim 12 is rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, because the specification, while being enabling for lactic acid bacterial cells modified to reduce **LDH** activity by **at least 10%** with treatment under aerobic conditions, does not reasonably provide enablement for lactic acid bacterial cells modified when fermented under **anaerobic** conditions. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make the invention commensurate in scope with these claims. To produce such cells to the full extent of their scope would require undue experimentation.

The factors to be considered in determining whether undue experimentation is required are summarized above.

In the instant application, *L. lactis* CHCC373 cells are treated with 10 mg/L haemin and grown in anaerobic or aerobic conditions. The cell extract (supernatant) and pellet (debris) were assayed. The assay performed shows 17% reduced LDH activity with haemin treatment under aerobic conditions in the cell extract, i.e. associated with the cellular material, and only 5% reduction with haemin treatment under anaerobic conditions in the cell extract. Thus, the experiment described does not support the claim to cells grown anaerobically that can reduce LDH activity by at least 10%. While minimal guidance in the use of other porphyrin-containing compounds is offered, no other working examples to help define the scope of the claims are described. The state of the prior art is such that numerous porphyrin compounds are known, but

all have different LDH activity with different bacteria under different aeration conditions. Therefore, it is wholly unpredictable how to achieve a cell that reduces LDH activity by at least 10% when cells have undergone treatment with haemin under anaerobic conditions.

28. Claim 16 is rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, because the specification, while being enabling for lactic acid bacterial cells modified to contain at least 0.1 ppm cytochrome d using mixed lactic acid bacterial strains grown aerobically, does not reasonably provide enablement for lactic acid bacterial cells modified to contain at least 0.1 ppm of **any porphyrin** containing compound **using pure strains**. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make the invention commensurate in scope with these claims. To produce such cells to the full extent of their scope would require undue experimentation.

The factors to be considered in determining whether undue experimentation is required are summarized above.

In the instant application, cells of a mixed strain of *Lactococcus lactis* are treated with 10 mg/L haemin and grown in anaerobic or aerobic conditions. The cell extract (supernatant) was assayed. The assay performed identified cytochrome d in the supernatant of only the aerobically grown cells (not anaerobically grown) and estimated cytochrome d to be present at a concentration of 13 ppm. Thus, 10 mg/L haemin-treated *L. lactis* mixed cells have cytochrome d within their cells when cells are fermented under aerobic conditions. No guidance as to how to produce such a result under anaerobic treatment conditions is offered with mixed strains. No guidance as to the haemin vs. cytochrome d is offered with mixed strains. No other working examples to help define the scope of the claims are described. The state of the prior art is such

that numerous porphyrin compounds are known, but all have different cellular association characteristics with different bacteria. Therefore, it is wholly unpredictable how to achieve the containment of at least 0.1 ppm of any porphyrin compound (not just cytochrome d) under anaerobic conditions and/or under mixed strain culture conditions.

29. Claims 40-42 are rejected under 35 U.S.C. § 112, first paragraph, enablement, as failing to comply with the enablement requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention at "at least **60 ppm**" (emphasis added) or higher of a **porphyrin-containing compound**. To produce such cells to the full extent of their scope would require undue experimentation.

The factors to be considered in determining whether undue experimentation is required are summarized above.

In the instant application, *L. lactis* CHCC373 cells are treated with 10 mg/L haemin and grown in anaerobic or aerobic conditions. The cell extract (supernatant) and pellet (debris) were assayed. The assay performed shows 41 ppm haemin found in the cell debris, i.e. associated with the cellular material, and not in the cell extract when cells are grown under anaerobic or aerobic conditions. Thus, 10 mg/L haemin-treated *L. lactis* cells retain 41 ppm haemin attached to their cellular material when cells are fermented under aerobic or anaerobic conditions. While minimal guidance in the use of porphyrin-containing compounds is offered to produce more porphyrin within the cells, no working examples to help define the scope of the claims are described. The state of the prior art is such that numerous porphyrin compounds are known, but all have different cellular association characteristics with different bacteria. Therefore, it is

wholly unpredictable how to achieve the containment of at least 60 ppm of a porphyrin-containing compound.

30. Claims 45-47 are rejected under 35 U.S.C. § 112, first paragraph, enablement, as failing to comply with the enablement requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention at "at least **40 ppm**" (emphasis added) or higher of a **cytochrome**. To produce such cells to the full extent of their scope would require undue experimentation.

The factors to be considered in determining whether undue experimentation is required are summarized above.

In the instant application, a mixed strain of *Lactococcus lactis* are treated with 10 mg/L haemin and grown in anaerobic or aerobic conditions. The cell extract (supernatant) was assayed. The assay performed identified cytochrome d in the supernatant of only the aerobically grown cells (not anaerobically grown) estimates cytochrome d to be present at a concentration of 13 ppm. Thus, 10 mg/L haemin-treated *L. lactis* mixed cells have 13 ppm cytochrome d within their cells, but not attached to their cellular material, when cells are fermented under aerobic conditions. No working examples of at least 40 ppm cytochrome content are described. The state of the prior art is such that numerous porphyrin compounds are known, but all have different cellular association characteristics with different bacteria. Therefore, it is wholly unpredictable how to achieve the containment of at least 40 ppm of cytochrome d.

31. Claims 48-52 are rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, because the specification, while being enabling for cells that, after having been treated with 10 mg/L haemin and inoculated into milk can reduce the amount of dissolved oxygen by about 35% per hour, does not reasonably provide enablement for cells otherwise treated that reduce the amount of dissolved oxygen **at greater than 35% per hour**. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make the invention commensurate in scope with these claims. To produce such cells to the full extent of their scope would require undue experimentation.

The factors to be considered in determining whether undue experimentation is required are summarized above.

In the instant application, *L. lactis* CHCC373 cells are treated with 10 mg/L haemin and grown in anaerobic or aerobic conditions. Whole cells where then used to inoculate milk and reduce the dissolved oxygen in fermentation at 30°C for 2 hours. Anaerobic cells removed 41% and 43% (+/- 10 mg/L haemin) of the dissolved oxygen in 2 hrs; aerobic cells removed 57% and 69% (+/- 10 mg/L haemin) of the dissolved oxygen in 2 hrs. Thus, the experiment described supports claims to up to 35% per hour (half of 69%) when the appropriate pre-treatment in haemin and aerobic conditions are noted. While minimal guidance in the use of other porphyrin-containing compounds is offered, no other working examples to help define the scope of the claims are described. The state of the prior art is such that numerous porphyrin compounds are known, but all have different oxygen consumption characteristics with different bacteria. Therefore, it is wholly unpredictable how to achieve a cell that consumes at least 40% of the dissolved oxygen. It is also wholly unpredictable how to reduce oxygen in media other than that

which is describes (milk at 30°C for 2 hours), particularly since the porphyrin-treatment effect is wholly uncharacterized, as far as structure and function of the treated cells, in the lactic acid bacteria.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. § 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

32. Claims 1, 4-7, 10-17, 35-39, 43, 44, 48, and 49 are rejected under 35 U.S.C. § 103(a) as being unpatentable over Kaneko *et al.* (USPN 5,075,226 – see IDS Paper No. 6). The instant claims are drawn to *Lactococcus lactis* cells that have been treated with 10 mg/L haemin under aerobic conditions. The instant claims are also drawn to starter cultures of said cells in frozen liquid or freeze-dried forms with cryoprotectant having a viability of 10^4 to 10^{12} CFU per gram. The instant claims are also drawn to mixed cultures of the same. All other limitations in the instant claims are inherent based on the treatment with 10 mg/L haemin that is disclosed in the examples of the instant specification.

Kaneko *et al.* (USPN 5,075,226) teach methods of culturing lactic acid bacteria, aerobically, in hemin (MW 563 g/mol) at a concentration of 0.1-500 μM (see examples and Claim 3); 500 μM is equivalent to approximately 327 mg/L hemin. Kaneko *et al.* (USPN 5,075,226) teach the use of *Lactococcus lactis* from the ATCC (see column 3, lines 60-65), which cells are delivered either as frozen liquid or freeze dried with a cryoprotectant added.

Kaneko *et al.* (USPN 5,075,226) teach using concentrations of 1-2% in inoculations as a starter culture (see column 6, examples 2 and 4), which concentration exceeds the requirement of 10^{12} CFU. While Kaneko *et al.* (USPN 5,075,226) describe methods of greater than 325 mg/L hemin, only as high as 3 mg/L hemin treatment was performed (see Comparative Example, column 5).

It would have been obvious to one of ordinary skill in the art to produce the claimed cells because said cells are within the scope of the claimed invention of Kaneko *et al.* (USPN 5,075,226). One would have been motivated to practice the full scope of the invention of Kaneko *et al.* (USPN 5,075,226) for the purpose of effectively producing diacetyl and acetoin, which are commercially useful flavoring agents. One would have had a reasonable expectation of success that the treatment of 10 mg/L hemin would produce the desired effect in the method described by Kaneko *et al.* (USPN 5,075,226) because no deleterious effects of high hemin concentrations are noted. All other characteristics in the claims, which are enabled by the specification by virtue of its limited examples, would naturally be found in *L. lactis* cells treated with 10 mg/L hemin.

Summary of Pending Issues

33. The following is a summary of the issues pending in the instant application:

a) The specification stands objected to for being confusing concerning porphyrin content throughout the examples presented.
b) Claim 14 stands objected to for having improper punctuation.
c) Claims 8 and 9 stand rejected under 35 U.S.C. § 112, second paragraph, as being indefinite for the term "about".
d) Claim 34 stands rejected under 35 U.S.C. § 112, second paragraph, as being indefinite for the unclear treatment of DSM12015.
e) Claim 34 stands rejected under 35 U.S.C. § 112, first paragraph, enabling deposit.
f) Claims 1, 4-17, 34-52 stand rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, related to containing at least 0.1 ppm of any porphyrin containing compound.

g) Claim 5 stands rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, relating to containing at least 0.1 ppm cytochrome d when fermented under anaerobic conditions
h) Claims 8-9 stand rejected under 35 U.S.C. § 112, first paragraph, enablement, relating to being inoculated at a concentration of 10^7 cells/ml into low pasteurised skimmed milk having 8 ppm of dissolved oxygen.
i) Claim 9 stands rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, relating to consuming at least 50% of dissolved oxygen with treatment under anaerobic conditions
j) Claim 11 stands rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, relating to reducing NOX activity by at least 10% with treatment under anaerobic conditions.
k) Claim 12 stands rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, relating reducing LDH activity by at least 10% with treatment under anaerobic conditions.
l) Claim 16 stands rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, relating to containing at least 0.1 ppm cytochrome d using pure strains.
m) Claims 40-42 stand rejected under 35 U.S.C. § 112, first paragraph, enablement, relating to containing at least 60 ppm or higher of a porphyrin-containing compound.
n) Claims 45-47 stand rejected under 35 U.S.C. § 112, first paragraph, enablement, relating to containing at least 40 ppm or higher of a cytochrome.
o) Claims 48-52 stand rejected under 35 U.S.C. § 112, first paragraph, scope of enablement, relating to reducing the amount of dissolved oxygen at greater than 35% per hour.
p) Claims 1, 4-7, 10-17, 35-39, 43, 44, 48, and 49 stand rejected under 35 U.S.C. § 103(a) as being unpatentable over Kaneko *et al.* (USPN 5,075,226).

Conclusion

34. Claims 1, 4-17, and 34-52 are not allowed for the reasons identified in the numbered sections of this Office action. Applicants must respond to the objections/rejections in each of the numbered sections in this Office action to be fully responsive in prosecution. The instant Office action is **NON-FINAL** based on newly set forth enablement and art rejections.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Kathleen M Kerr whose telephone number is (703) 305-1229. The examiner can normally be reached on Monday through Friday, from 8:30am to 5pm.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Ponnathupura Achutamurthy can be reached on (703) 308-3804.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is (703) 308-0196.

KMK
July 31, 2003